CLAIMS

1. An isolated enzyme capable of mediating a site-specific recombination between two predetermined recombination sites, wherein at least one recombination site is an asymmetric recombination site.

2. The isolated enzyme according to claim 1, wherein the recombination is selected from a group consisting of: inversion of a first DNA molecule encompassed within a second DNA molecule, excision of a first DNA molecule from a second DNA molecule, insertion of a first DNA molecule into a second DNA molecule and translocation between a first DNA molecule and a second DNA molecule.

3. The isolated enzyme according to claim 2, wherein the second DNA molecule is selected from the group consisting of: genomic DNA and circular DNA.

4. The isolated enzyme according to claim 2, wherein the second DNA molecule is genomic DNA and the first DNA molecule is integrated into a predetermined genomic site selected from the group consisting of: 3' UTRs, 5' UTRs, polyA sites and gene promoters.

5. The isolated enzyme according to claim 1, wherein said isolated enzyme is a Cre or FLP mutant mediating recombination between two recombination sites, such that at least one recombination site is an asymmetric recombination site comprising a spacer sequence selected from the group consisting of: SEQ ID NOS. 1-34.

6. A plurality of isolated enzymes capable of mediating site-specific recombination between two predetermined recombination sites, wherein at least one of the recombination sites is an asymmetric recombination site.

7. The plurality of isolated enzymes according to claim 6, wherein at least one enzyme is a wild type recombinase.

8. The plurality of isolated enzymes according to claim 6, wherein at least one enzyme is a Cre or Flp mutant mediating recombination

between two recombination sites, such that at least one recombination site is an asymmetric recombination site comprising a spacer sequence selected from the group consisting of: SEQ ID NOS. 1-34.

9. An isolated polynucleotide encoding an enzyme, the enzyme is capable of mediating site-specific recombination between two recombination sites, wherein at least one of the recombination sites is an asymmetric recombination site.

10. The isolated polynucleotide according to claim 9, wherein said isolated polynucleotide is encompassed in a recombinant vector that expresses the at least one recombinase.

11. The isolated polynucleotide according to claim 10, wherein the recombinant vector is selected from the group consisting of: naked DNA plasmid, a plasmid within a liposome, a retroviral vector, an AAV vector, or a recombinant adenoviral vector.

12. The isolated polynucleotide according to claim 10, wherein the recombinant vector further comprising a promoter.

13. The isolated polynucleotide according to claim 12, wherein the promoter is derived from bacteria, yeast, insect, animal, plant and virus.

14. The isolated polynucleotide according to claim 12, wherein the promoter is selected from the group consisting of: E. coli *lac* and *trp* operons, the *tac* promoter, the bacteriophage$_{\lambda}$L promoter, bacteriophage T7 and SP6 promoters, β-actin promoter, insulin promoter, human cytomegalovirus (CMV) promoter, HIV-LTR, RSV-LTR, SV40 promoter, baculoviral polyhedrin and p10 promoter.

15. The isolated polynucleotide according to claim 12, wherein the promoter is an inducible promoter.

16. The isolated polynucleotide according to claim 15, wherein the inducible promoter is selected from the group consisting of: tetracycline, heat shock, steroid hormone, heavy metal, phorbol ester, adenovirus E1A element, interferon and serum inducible promoters.

17. The isolated polynucleotide according to claim 9, wherein said isolated polynucleotide encodes a plurality of enzymes, the plurality of enzymes is capable of mediating site-specific recombination between two predetermined recombination sites, wherein at least one of the recombination sites is an asymmetric recombination site.

18. The isolated polynucleotide according to claim 17, wherein each of the plurality of recombinases recognizes at least one half of the at least one asymmetric recombination site.

19. The isolated polynucleotide according to claim 17, wherein at least one recombinase is a Cre mutant mediating recombination between two recombination sites, such that at least one recombination site is an asymmetric recombination site comprising a spacer sequence selected from the group consisting of: SEQ ID NOS. 1-34.

20. A host cell comprising a vector, the vector encompassing polynucleotide encoding at least one enzyme, the at least one enzyme is capable of mediating site-specific recombination between two recombination sites, wherein at least one of the recombination sites is an asymmetric recombination site.

21. The host cell according to claim 20, capable of expressing said at least one enzyme.

22. A genetically modified cell transformed by an site-specific recombination between two recombination sites, wherein at least one of the recombination sites is an asymmetric recombination site, and wherein the asymmetric recombination is selected from the group consisting of: inversion, excision, insertion and translocation.

23. The genetically modified cell according to claim 22, wherein the recombination occurs between the cellular endogenous genome and an exogenous DNA molecule.

24. The genetically modified cell according to claim 22, wherein said genetically modified cell comprises an exogenous DNA molecule, wherein the exogenous DNA molecule is integrated by recombination

between two recombination sites, at least one of the recombination sites is an asymmetric recombination site, into a predetermined locus within the cellular genome.

25. The genetically modified cell according to claim 22, wherein said genetically modified cell is eukaryotic.

26. The genetically modified cell according to claim 22, wherein said genetically modified cell is selected from the group consisting of: yeast, plant cell, embryonic stem cell, mesenchymal cell, and haematopoietic progenitor cell.

27. A transgenic organism comprising the genetically modified cell of claim 22.

28. The transgenic organism according to claim 22, said transgenic organism is selected from the group consisting of: plant, yeast and mammal.

29. The genetically modified cell according to claim 22, wherein said cell is devoid of an endogenous polynucleotide sequence at a predetermined genomic locus.

30. The genetically modified cell according to claim 29, wherein said genetically modified cell is eukaryotic.

31. The genetically modified cell according to claim 29, wherein said genetically modified cell is selected from the group consisting of: yeast, plant cell, embryonic stem cell, mesenchymal cell, and haematopoietic progenitor cell.

32. A transgenic organism comprising the genetically modified cell of claim 29.

33. The transgenic organism according to claim 32, said transgenic organism is selected from the group consisting of: plant, yeast and mammal.

34. A method for treating a disease, comprising:

a. providing a composition comprising a DNA molecule comprising a nucleotide sequence encoding at least one enzyme, the at least one enzyme is capable of mediating site-specific excision of a gene fragment flanked between two recombination sites, wherein at least one recombination site is an asymmetric recombination site; and

b. administering the composition to a subject in need thereof.

35. The method according to claim 34, further comprising obtaining site-specific excision of the gene fragment from a predetermined genomic locus.

36. The method according to claim 34, wherein the composition further comprises a carrier operably connected to the isolated DNA molecule, the carrier capable of targeting said isolated DNA molecule to a cell.

37. The method according to claim 36, wherein the carrier promotes internalization of said isolated DNA molecule into the cell.

38. The method according to claim 36, wherein the carrier is selected from the group consisting of: viruses, liposomes, lipid/DNA complexes, micelles, protein/lipid complexes, nanoparticles, and microparticles.

39. The method according to claim 34, wherein the two recombination sites are the same asymmetric recombination sites.

40. The method according to claim 34, wherein the nucleotide sequence encodes a plurality of enzymes capable of catalyzing the recombination.

41. The method according to claim 34, wherein the excised gene fragment is a fragment of HIV genomic DNA.

42. The method according to claim 34, wherein the composition comprises a recombinant vector encompassing an expression cassette comprising the nucleotide sequence.

43. The method according to claim 42, wherein the vector is selected from the group consisting of: naked DNA plasmid, a plasmid within a liposome, retrovirus,

lentivirus, adenovirus, herpes simplex viruses (HSV), cytomegalovirus (CMV), and adeno-associated virus (AAV).

44. A method for mediating site-specific excision, comprising:

a. providing a composition comprising a DNA molecule comprising a nucleotide sequence encoding at least one enzyme, the at least one enzyme is capable of mediating site-specific excision of a gene fragment flanked between two recombination sites, wherein at least one recombination site is an asymmetric recombination site; and

b. transforming a cell with the composition.

45. The method according to claim 44, further comprising proliferating the transformed cells ex vivo.

46. The method according to claim 44, wherein the cell is autologous.

47. The method according to claim 44, further comprising obtaining site-specific excision of the gene fragment at a defined genomic locus within the cell.

48. The method according to claim 47, further comprising selecting cells devoid of said gene fragment.

49. The method according to claim 47, further comprising transplanting the selected cell into a subject in need thereof.

50. The method according to claim 45, wherein transforming the cell with said composition is carried out by a procedure selected from the group consisting of: calcium phosphate transfection, DEAE-dextran mediated transfection, transvection, microinjection, cationic lipid-mediated transfection, electroporation, scrape loading, ballistic introduction or infection, use of a gene gun and lyposome transfection.

51. The method according to claim 34, wherein the composition comprising at least one enzyme capable of mediating site-specific excision of the gene fragment flanked between two recombination sites, wherein at least one recombination site is an asymmetric recombination site.

52. The method according to claim 51, wherein the composition further comprising a carrier operably linked to said at least one enzyme, the carrier is capable of targeting said at least one enzyme to a specific cell.

53. The method according to claim 52, wherein the carrier further promotes internalization of said at least one enzyme into the specific cell.

54. A method for treating a disease, comprising:

a. providing a composition comprising a first DNA molecule comprising a first recombination site; and a second DNA molecule comprising a nucleotide sequence encoding at least one enzyme, the at least one enzyme mediates insertion of the first DNA molecule or fragments thereof into a third DNA molecule comprising a second recombination site; and

b. administering the composition to a subject in need thereof

wherein at least one of said first and second recombination sites is an asymmetric recombination site.

55. The method according to claim 54, wherein the first DNA molecule comprises a nucleotide sequence consisting of a fragment of human genomic DNA.

56. The method according to claim 54, wherein the first DNA molecule is a gene encoding for a molecule selected from the group consisting of: a structural protein, an enzyme and a regulatory molecule.

57. The method according to claim 54, wherein the third DNA molecule is genomic DNA.

58. The method according to claim 57, wherein the first DNA molecule is inserted into a defined locus of the genome selected from the group consisting of: 3' UTRs, 5' UTRs, polyA sites and gene promoters.

59. The method according to claim 54, wherein the composition further comprises a carrier operably connected to the first and second DNA molecules, the carrier capable of targeting said first and second DNA molecules to a cell encompassing the third DNA molecule.

60. The method according to claim 59, wherein the carrier is selected from the group consisting of: viruses, liposomes, lipid/DNA complexes, micelles, protein/lipid complexes, nanoparticles and microparticles.

61. The method according to claim 54, wherein the first DNA molecule and the second DNA molecule are operatively linked to one another.

62. The method according to claim 61, wherein the second DNA molecule is operably linked to a promoter.

63. The method according to claim 54, wherein the first DNA molecule comprises a recombination site comprising SEQ ID NO:37 and the second DNA molecule comprising a nucleotide sequence encoding at least one enzyme selected from the group consisting of: wild type Cre, CM1 Cre mutant and CM2 Cre mutant.

64. A method for mediating site-specific excision of a gene fragment, comprising:

a. providing a composition comprising a DNA molecule comprising a nucleotide sequence encoding at least one enzyme, the at least one enzyme is capable of mediating site-specific excision of a gene fragment flanked between two recombination sites, wherein at least one recombination site is an asymmetric recombination site; and

b. transforming a cell with the composition.

65. The method according to claim 64, further comprising proliferating the transformed cells ex vivo.

66. The method according to claim 64, wherein the cell is autologous.

67. The method according to claim 64, further comprising selecting cells comprising the first DNA molecule integrated within their genome at a predetermined locus.

68. The method according to claim 67, further comprising transplanting the selected cell into a subject in need thereof.

69. A method for mediating site-specific insertion, comprising:

a. providing a composition comprising

i. a first DNA molecule, the first DNA molecule comprises a first recombination site; and

ii. at least one enzyme capable of mediating site-specific insertion of the first DNA molecule into a second recombination site within a specific genomic locus; and

b. administering the composition to a subject in need thereof

wherein at least one of said first and second recombination site is an asymmetric recombination site.

70. The method according to claim 69, wherein the composition further comprising a carrier operably linked to said at least one enzyme, the carrier is capable of targeting said at least one enzyme to a specific cell encompassing the specific genomic locus and promoting internalization of said at least one enzyme into the specific cell.

71. The method according to claim 69, wherein the first DNA molecule comprises a recombination site comprising SEQ ID NO:37 and the at least one enzyme is selected from the group consisting of: wild type Cre, CM1 Cre mutant and CM2 Cre mutant.